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COMPARATIVE TOXICITY TESTS OF ANABASINE, NORNICOTINE, AND NICOTINE

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During the last 12 to 15 years the alkaloid anabasine, an isomer of nicotine, has been obtained by the Russians from Anabasis aphylla and the sulfate used commercially as an insecticide. Before the war small quantities of this material were imported into the United States. Other alkaloids common to A. aphylla, however, are also present in this product, so that the entomological tests that have been made on Russian anabasine sulfate are not reliable for this alkaloid unless further purification has been made. Anabasine is also obtainable from Nicotiana glauca Grah., a plant which grows wild in the southwestern United States.

Although there has been considerable entomological testing of nicotine and anabasine compounds, the paucity of results for nornicotine and the lack of comparative tests for these three alkaloids made it desirable that such testing be done.

Anabasine, nicotine, and nornicotine were therefore tested as low-concentration sprays (0.1 percent or less) in the laboratory at Anaheim, Calif., against the cabbage aphid (Brevicoryne brassicae (L.)), the celery leaf tier (Phlyctaenia rubigalis (Guen.)), the citrus red mite (Paratetranychus citri McG.), the large milkweed bug (Oncopeltus fasciatus (Dall.)), the nasturtium aphid (Aphis fabae Scop.), the pea aphid (Macrosiphum pisi (Kltb.)), and a spider mite (Tetranychus sp.).

Phytotoxicity tests of the three alkaloids were made under natural outdoor conditions by treating green foliage of pumpkins, beans, collards, peas, and turnips with a 0.2-percent solution of the alkaloid.

Sample Materials

The aqueous alkaloid solutions were prepared from nicotine, nornicotine, and anabasine that had been purified so that each alkaloid contained none of either of the others.

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The nicotine was freed of secondary amines by the formation of the nitroso derivatives and subsequent steam distillation from an alkaline solution.

The nornicotine was separated from nicotine according to the method of Smith (2), formed a picrate melting at 190-191° C. and on methylation (Bowen and Barthel, 1) yielded nicotine. The nornicotine was free of nicotine, as evidenced by the absence of alkaloids in the distillate after the formation of the nitroso compound.

The anabasine was obtained from Nicotiana glauca Grah., formed a picrate at 212-213° C., and after methylation formed a picrate melting at 236-237° C.

Test Insects

The aphids and mites used in these tests were collected either from natural infestations in the field or from populations propagated under outdoor cage conditions. The celery leaf tier larvae and milkweed bugs were reared under laboratory conditions. Both nymphal and adult stages of the mites and aphids, except pea aphids, were used for testing. Only 2-day-old nymphs of the pea aphid were tested, and fourth instars of the milkweed bug and the celery leaf tier.

Methods

Aphids and mites were sprayed while present on their host plant. Plants for aphid hosts were grown in small terra cotta pots, and were infested only a few days before the tests were made. For the tests with mites, leaves infested naturally under outdoor conditions were used, the stems being placed in 120-ml. flasks of water. After treatment the pots and flasks were placed in battery jars approximately 6 inches wide by 8 inches high, which were then covered with muslin stretched tightly across their top and held in place by a rubber band. The mortality counts were made 2 days after treatment in tests against all insects except the celery leaf tier; a 3-day test period was allowed for this insect.

Celery leaf tier larvae were confined in petri dishes with sections of treated swiss chard leaves.

The milkweed bugs were sprayed thoroughly and then confined with untreated food (milkweed seed) in crystallizing dishes with a petroleum-jelly barrier around the rim to prevent escape of the bugs.

Host materials were treated thoroughly with a fine spray of the aqueous mixtures released under approximately 10 pounds' pressure by an atomizer-type spray gun.

Anabasine at a concentration of 0.025 percent was more effective than nicotine at the same concentration against the cabbage aphid and pea aphid. However, against the cabbage aphid the difference was not significant. At a concentration of 0.0125 percent anabasine was more effective than nicotine against the nasturtium aphid, although the difference was not quite enough to be significant. At a concentration of 0.1 percent against the celery leaf tier, the citrus red mite, the milkweed bug, and the common red spider, only one significant difference in treatments appeared. Results from the anabasine treatment against citrus red mites were significantly higher than from the nicotine treatment.

At the concentrations used nornicotine was less toxic than either nicotine or anabasine to the cabbage aphid, the celery leaf tier, the large milkweed bug, and the red spider; it was slightly more toxic than nicotine, but less toxic than anabasine, to the citrus red mite, the nasturtium aphid, and the pea aphid. However, none of these differences were significant.

No apparent injury to green foliage resulted from these treatments.

All three samples were tested simultaneously against each insect. Tests were replicated 5 times against all insects except the citrus red mite. In this test only 4 replicates were made. The data were subjected to an analysis of variance.

Phytotoxicity tests were made by spraying two plants with a small hand sprayer, the leaves being sprayed thoroughly on both sides. Two applications were made 7 days apart. Observations to detect injury were made 8 and 15 days after the first treatment.

Results

The results of these tests are summarized in table 1.

Table 1.--Effectiveness of anabasine, nicotine, and nornicotine in different concentrations for the control of various insects.

Insect	Concentration Percent	Material	Insects Tested Number	Insect Mortality Percent
Cabbage aphid	0.025	Anabasine	222	92.0
		Nicotine	118	78.0
		Nornicotine	186	74.0
		Check	161	1.2
Difference required for significance, 5% level				20.5
Celery leaf tier	1.0	Nicotine	40	40.4
		Anabasine	40	32.8
		Nornicotine	40	22.6
		Check	40	0.0
Difference required for significance, 5% level				28.2
Citrus red mite	1.0	Anabasine	117	56.2
		Nornicotine	101	26.6
		Nicotine	85	24.8
		Check	96	2.1
Difference required for significance, 5% level				30.7
Large milkweed bug	1.0	Nicotine	61	96.4
		Anabasine	75	79.9
		Nornicotine	63	61.6
		Check	52	0.0
Difference required for significance, 5% level				36.3
Nasturtium aphid	0.0125	Anabasine	194	94.8
		Nornicotine	253	73.8
		Nicotine	284	54.2
		Check	337	4.2
Difference required for significance, 5% level				41.2
Pea aphid	0.025	Anabasine	157	100.0
		Nornicotine	136	90.8
		Nicotine	163	69.2
		Check	74	1.4
Difference required for significance, 5% level				27.3
Red spider	1.0	Nicotine	573	28.6
		Anabasine	543	22.6
		Nornicotine	542	21.0
		Check	125	4.0
Difference required for significance, 5% level				27.9

Literature Cited

(1) Bowen, C. V., and W. F. Barthel. 1944. Identification of nornicotine in tobacco. Indus. and Engin. Chem., Analyt. Ed. 16 (6): 377-8.

(2) Smith, C. R. 1942. Azeotropism in the system nicotine-water. Separation of nicotine from related alkaloids by aqueous distillation. Indus. and Engin. Chem., Indus. Ed. 34 (2): 251-2.

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